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Association between urinary 3, 5, 6-trichloro-2-pyridinol, a metabolite of chlorpyrifos and chlorpyrifos-methyl, and serum T4 and TSH in NHANES 1999–2002

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Abstract

Thyroid hormones are vital to a host of human physiological functions in both children and adults. Exposures to chemicals, including chlorpyrifos, have been found to modify thyroid signaling at environmentally relevant levels in animal studies. The aim of this study was to examine circulating T4 and TSH levels in relation to urinary concentrations of 3, 5, 6-trichloro-2-pyridinol (TCPY), a metabolite of the organophosphorus insecticides chlorpyrifos and chlorpyrifos-methyl, using data from individuals 12 years and older from the U.S. National Health and Nutrition Examination Surveys (NHANES). NHANES datasets from 1999–2000 and 2001–2002 were combined, and individuals with thyroid disease, those taking thyroid medications, and pregnant women were excluded (N=3249). Multivariable linear regression models for relationships between log-transformed urinary TCPY and serum total T4 or log (TSH) were constructed adjusting for important covariates. Models were stratified by sex and a categorical age variable (12–18, 18–40, 40–60, and >60 years). In male participants, an interquartile range (IQR) increase in urinary TCPY was associated with statistically significant increases in serum T4 of 3.8% (95th CI 0.75 to 7.0) among those 12–18 years of age and 3.5% (95th CI 0.13 to 7.0) in the 18–40 year age group, relative to median T4 levels using unweighted models. An IQR increase in TCPY was also associated with decreases in TSH of 10.7% (−18.7 to −2.05) among men 18–40 years old and 20.0% (95th CI −28.9 to −9.86) among men >60 years old. Conversely, urinary TCPY was positively associated with TSH in females >60 years of age. Further research to confirm these findings, elucidate mechanisms of action, and explore the clinical and public health significance of such alterations in thyroid hormones is needed.

Keywords

Biomarker; Endocrine Disruption; Exposure; Pesticides; Human
1. INTRODUCTION

Globally and domestically, chlorpyrifos is the most widely used non-persistent, organophosphate (OP) pesticide (Ye et al., 2008; Bradman and Whyatt, 2005; Timchalk et al., 2007) with approximately 10 million pounds agriculturally applied annually in the U.S. (EPA, 2011). Residential use of chlorpyrifos in the U.S. was banned in 2000, but widespread exposure remains likely in the U.S. and elsewhere (EPA, 2001). Environmental human exposure is ubiquitous and occurs through inhalation of vapors and aerosols from spray drift (Pang et al., 2002; Whyatt et al., 2009), ingestion of residuals on food and house dust/soil (Pang et al., 2002; Salas et al., 2003), and dermal absorption following skin contact (Panuwet et al., 2008) and is usually excreted within hours or days in urine (Bradman and Whyatt, 2005; Timchalk et al., 2007). Human exposure has been quantified from various media such as indoor air (Pang et al., 2002), carpet dust (Pang et al., 2002), breast milk (Weldon et al., 2011), fruit (Riederer et al., 2009), and water (Carvalho et al., 2002). Human exposure has not only been quantified in the U.S. but in other countries such as The Netherlands (Ye et al., 2008), China (Panuwet et al., 2008), Germany (Koch et al., 2001), and Italy (Aprea et al., 1999).

Environmental exposures to chemicals have been found to modify thyroid hormone signaling (Blount et al., 2006; De Angelis et al., 2009), which are vital to a host of physiological functions in both children and adults (Yen et al., 2001; Thrasher et al., 2002), even at low-levels of exposure. Studies examining thyroid hormones in relation to non-persistent pesticides remain limited but animal and human studies suggest that chlorpyrifos or other organophosphate pesticides may alter thyroid hormone levels (Lacasaa et al., 2010; Meeker et al., 2006). In animal studies, decreased T4 and cellular changes within the thyroid and pituitary glands in response to chlorpyrifos exposure were observed (Ghisari and Bonefeld-Jorgensen, 2005; De Angelis et al., 2009; Jeong et al., 2006).

In the present study, we assessed the relationship between urinary TCPY, a biomarker of exposure to chlorpyrifos, chlorpyrifos-methyl, or TCPY, and altered serum thyroid hormone levels using data from the U.S. National Health and Nutrition Examination Survey (NHANES).

2. METHODS

2.1 Study Population

The National Health and Nutrition Examination Survey (NHANES) is a cross-sectional study designed to be representative of the civilian, non-institutionalized population of the United States. Sample design consists of a stratified, multistage, probability sample of civilian, non-institutionalized adults and children (CDC, 2010). A laboratory subsample (one-third of the total sample) in which both urinary TCPY and thyroid measurements were taken for the 1999–2000 and 2001–2002 NHANES surveys were used for this analysis. This subsample consisted of 3249 individuals after the exclusion of pregnant women (N=194), participants taking thyroid (and antithyroid) medications including levothyroxine sodium (N=103), and those who currently have or have ever had thyroid disease (N=141).

2.2 Measurements

Sociodemographic information obtained from NHANES demographic files included sex, race, household income, and age. Body mass index (BMI) (kg/m^2) was obtained from the body measures examination data files. Data on urinary 3, 5, 6-trichloro-2-pyridinol (TCPY), urinary creatinine, serum total thyroxine (T4) and thyrotropin (TSH), and serum cotinine were obtained from the NHANES laboratory data files.
Medications were identified in the prescription drug medication and the analgesics/pain reliever questionnaires and included: estrogens and/or progesterone, other steroid hormones (androgens, adrenal corticosteroids, tamoxifen, raloxifene, and pituitary hormones), non-steroidal anti-inflammatory drugs (NSAIDs), furosemide, beta-blockers, blood glucose regulators, and other medications thought to affect thyroid hormones (amiodarone, carbamazepine, chlorpropamide, carbidopa/levodopa, heparin, interferon, lithium, phenytoin, phenobarbital, and sulfasalazine) (Turyk et al. 2007).

2.3 Laboratory Methods

**Total thyroxine**—Samples collected in 1999–2000 and 2001 were analyzed for serum T\textsubscript{4} using the Hitachi 704 method while in 2002 T\textsubscript{4} levels in serum samples were determined using a paramagnetic particle, chemiluminescent, competitive binding enzyme immunoassay on a Beckman Access\textsuperscript{2} Immunoassay System (Aoki et al., 2007; CDC, 2006). The analytical range for T\textsubscript{4} was 6.4 to 390 nmol/L (0.5 – 30.3 μg/dL) and the laboratory reference (i.e., normal) range was 69.5 to 164.7 nmol/L (5.4 to 12.8 μg/dL) (Aoki et al., 2007; CDC, 2006).

**Thyrotropin**—1999–2000 and 2001 samples were analyzed for TSH using the IMx ultrasensitive hTSH II microparticle enzyme immunoassay technique while in 2002 a two-site, paramagnetic particle chemiluminescent immunoassay was used. Analytical sensitivity ranged from 0.01 to 100 mIU/L (Aoki et al., 2007); (CDC, 2006). The laboratory reference range for 1999–2000 and 2001 samples were reported as 0.47 to 5.01 mIU/L (Aoki et al., 2007; CDC, 2006). The laboratory reference range for 2002 samples was reported as 0.24 to 5.4 mIU/L (Aoki et al., 2007; CDC, 2006).

**TCPY**—Urine samples (2 mL) were spiked with stable isotopically labeled TCPy and then subjected to an enzyme hydrolysis to liberate glucuronide- and sulfate-bound TCPy. Hydrolysates were extracted using mixed-polarity solid-phase extraction cartridges (CDC, 2006; Olsson et al., 2004). Elutes (methanol) were concentrated and analyzed using HPLC/tandem MS (Olsson et al., 2004) with both quantification and confirmation ions monitored. TCPY was quantified using isotope dilution calibration. The limit of detection (LOD) of TCPY was 0.40 μg/L in urine.

**Creatinine**—Urine samples were analyzed via the Jaffé rate reaction, in which creatinine reacts with picrate in an alkaline solution to form a red creatinine-picrate complex, using a Beckman CX3 Chemistry analyzer (CDC, 2006; Barr et al 2005).

2.4 Statistical Analysis

The Survey Procedures in SAS Version 9.2 was utilized for most analyses. Descriptive statistics for demographic information were calculated along with the distribution of TCPY and thyroid hormones. Values equal to the limit of detection (LOD) (0.40 μg/L) divided by the square root of two were imputed for values of TCPY less than the LOD. Appropriate statistical weights were used to adjust for study design, oversampling and non-response. However, we constructed models both with and without including the sample weights since the weighted method may result in an inefficient analysis due to the large variability in assigned weights, as well as when covariates used in the creation of weights (such as age, sex, and ethnicity) are included in the analysis (Korn and Graubard, 1991).

Because some variables were not normally distributed, we used natural log (ln)-transformations of TSH and TCPY for analysis while serum T\textsubscript{4} was modeled untransformed. Bivariate analyses between dependent variables serum T\textsubscript{4} and log (TSH) and the independent variable log (TCPY) were conducted as well. Multivariable regression was used to construct separate models for each hormone stratified by gender and a
categorical age variable using PROC SURVEYREG. Age was categorized as follows: 12–<18yrs, 18–40yrs, 40–60yrs, and >60yrs. Continuous variables for age, BMI, serum cotinine, and (log) urinary creatinine, and categorical variables for race and income, were included in adjusted models. Race was categorized as: Mexican Americans, Whites, African Americans, and Other. Income was categorized as: less than $19,999 per year and greater than $20,000 per year. We also considered categories for prescribed thyroid medications: Furosamide, Betablockers, NSAID, Steroids, Other drug, estrogen (E2), and Estrogen/Progesterone (E2prog) medications. However, their inclusion did not impact effect estimates and they were not included in final models. To improve interpretability, regression coefficients were presented as a percent change in serum T4 and TSH for an interquartile range (IQR) increase in urinary TCPY levels. Interaction terms for TCPY with sex and age were explored in a secondary analysis. Lastly, using logistic regression models, we explored whether TCPY was associated with being categorized as hypothyroid or hyperthyroid based on laboratory reference ranges.

3. RESULTS

Descriptive statistics are shown in Table 1. In this population, 16% of TCPY measurements were below the LOD. Table 2 shows results for crude and adjusted regression coefficients for associations between urinary TCPY and serum T4 and TSH, stratified by sex. Results of the crude analyses revealed a positive relationship between urinary TCPY and serum T4 that was significant for males between the ages of 18 – 40 years for the both statistically weighted and unweighted models. In the unweighted multivariable model for males 18 – 40 years of age, an increase of 3.54% (95th CI 0.13 to 6.96) was observed for serum T4 levels in relation to an interquartile range (IQR) increase in TCPY levels. For males 12–<18 years of age, an IQR increase in TCPY was associated with a 3.85% (95th CI 0.75 to 6.95) increase in serum T4. In males >60 years of age an IQR increase in TCPY was associated with a 14.5% (95th CI −27.6 to −1.11) decrease in TSH. Conversely, TCPY was positively associated with TSH in women >60 years of age. Effect estimates in the weighted multivariable models were overall similar to those from the unweighted models. An IQR increase in TCPY was associated with 10.7% (95th CI −18.7 to −2.05) 20.0% (95th CI −28.9 to −9.86) decreases in TSH, among males 18 – 40 and >60 years of age, respectively.

In our secondary analysis (results not shown) the interaction term between TCPY and age was statistically significant (p<0.05), and the interaction term between TCPY and sex statistically suggestive (p<0.1), in relation to serum T4 in multivariable models. Neither of these interaction terms approached statistical significance for TSH. Finally, no relationship was observed between urinary TCPY and hypothyroid or hyperthyroid status.

4. DISCUSSION

The results of this study suggest that there is a positive relationship between urinary TCPY and serum total T4, and a negative relationship between TCPY and serum TSH, in adolescent males and/or men of reproductive age. There was also evidence of decreased and increased TSH in relation to urinary TCPY among males and females >60 years of age, respectively. These findings add to the existing evidence that exposure to certain organophosphate insecticides or their metabolites may disrupt the hypothalamic-pituitary-thyroid (HPT) axis. However, the exact mechanism of action is not understood, as there are only limited reports to date regarding the relationship between exposure to chlorpyrifos or chlorpyrifos-methyl and thyroid function.

In a human observational study, an inverse association between urinary TCPY and free T4 was reported in 322 adult men of reproductive age recruited through an infertility clinic (Meeker et al., 2006). This was not consistent with results from the present study, where we
observed a positive association between urinary TCPY and total T4 in adolescent males (<18 years) and males 18–40 years of age. However, serum levels of free T4 were not available in NHANES 1999–2002, and the study of men from an infertility clinic did not measure total T4. In an occupational study of 136 male floriculture workers that examined the association between thyroid hormones (T3, T4, and TSH) and OP exposure, urinary dialkyl phosphate (DAP) concentrations were associated with increased levels of TSH and total T4 which supports the results of this study; however, urinary DAPs may reflect exposure to numerous OPs and exposure to chlorpyrifos more specifically was not assessed in the study (Lacasana et al., 2010).

Several animal studies investigating thyroid effects related to chlorpyrifos or other OPs may support our results suggesting that TCPY alters thyroid signaling, though the specific findings have not fully consistent between studies. Our observation of sex differences in these relationships is supported by a couple of these studies, whereas experimental support or explanation for the differences we found related to age is lacking. In a study involving male Wistar albino rats subjected to acute organophosphate (methamidophos; dimethyl phosphoramidothioate) exposure, a decrease in serum T4, T3, and TSH levels was observed and resulted in secondary hypothyroidism and sick euthyroidism (Satar et al 2005). A decrease in serum T4 levels was also observed in CD1 mice (both in dams and F1) after developmental exposure to chlorpyrifos at doses low enough to not elicit inhibition of brain acetylcholinesterase (AchE) (De Angelis et al., 2009). Both sexes of F1 CD1 mice showed reduced serum T4 levels, and, perhaps consistent with the present study, a more significant effect was observed in males compared to females (De Angelis et al., 2009). Jeong et al. (2006) reported that chlorpyrifos-methyl induces hypothyroidism (decreased serumT4 and increased serum TSH) and altered thyroid and pituitary gland weights through sexual maturation and adulthood in rats after long-term in utero and postnatal exposure (Jeong et al., 2006). Interestingly, dose-response relationships between exposure and thyroid hormones appeared stronger among the male rates. Finally, the thyroid disrupting potential of chlorpyrifos was also demonstrated in vitro by a study of rat pituitary GH3 cells, where chlorpyrifos exposure altered T3-induced cell growth (Ghisari and Bonefeld-Jorgensen, 2005).

Our analysis had several limitations. The analysis was based on a single measure of urinary TCPY and serum thyroid hormone levels. Despite the short half-life of chlorpyrifos (approximately 27 hours in the body) and substantial temporal variability in exposure levels over time (CDC, 2010; Meeker et al. 2005), urinary TCPY is still considered to be the best and most specific biomarker of chlorpyrifos and chlorpyrifos-methyl exposure (Barr et al., 2006). Thyroid hormone levels, especially TSH, may also have significant intraindividual variability over time (Hollowell et al., 2002; Surks et al., 2005). Another limitation is that pregnant women, infants, and young children were not included in this analysis. Human and experimental studies have shown that fetuses and infants were more sensitive than adults to many environmental toxicants, including chlorpyrifos (Timchalk et al., 2007). Also, there is growing evidence of the adverse impact of exposure to chlorpyrifos on fetal growth and early childhood neurodevelopment (Perera et al., 2005; Eskenazi et al., 2007; Rauh et al., 2006, 2011; Engel et al., 2011). Nevertheless, our findings of a relationship between urinary TCPY and markers of thyroid function may help inform potential mechanisms involved in adverse childhood neurodevelopment associated with OP exposure but should be explored in developmental cohort studies.

Despite the above limitations, this is the largest study to date examining the relationship between thyroid hormones and a biomarker of chlorpyrifos exposure in a human population. This will add to the body of knowledge assessing how environmental exposures impact thyroid signaling, which is vital to numerous human physiologic functions.
Acknowledgments

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Abbreviations

<table>
<thead>
<tr>
<th>TH</th>
<th>Thyroid Hormone</th>
</tr>
</thead>
<tbody>
<tr>
<td>TCPY</td>
<td>3, 5, 6-trichloro-2-pyridinol</td>
</tr>
<tr>
<td>NHANES</td>
<td>National Health and Nutrition Examination Surveys</td>
</tr>
<tr>
<td>T4</td>
<td>Thyroxine</td>
</tr>
<tr>
<td>TSH</td>
<td>Thyrotropin or Thyroid Stimulating Hormone</td>
</tr>
<tr>
<td>LOD</td>
<td>Limit of Detection</td>
</tr>
</tbody>
</table>

References


Engel SM, Wetmur J, Chen J, Zhe C, Barr DB, Canfield RL, Wolff MS. Prenatal exposure to organophosphates, paraoxonase 1, and cognitive development in childhood. Environ Health Perspect. 2011; 119(8)


Highlights

- Urinary TCPY is a biomarker of the common organophosphate insecticide chlorpyrifos
- TCPY was associated with higher T4 among males aged 12–<18 and 18–<40 years of age
- TCPY also associated with a suggestive decrease in TSH among men 18–<40 years old
- Contradicting associations were found for TSH among men and women >60 years old
Table I


<table>
<thead>
<tr>
<th>Variable</th>
<th>12 – &lt;18 years (n=506; n=550)</th>
<th>18–40 years (n=506; n=511)</th>
<th>40–60 years (n=377; n=333)</th>
<th>&gt;60 years (n=200; n=218)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td><strong>Age (yrs)</strong></td>
<td></td>
<td></td>
<td>15.0(13.0, 16.0)</td>
<td>14.0(13.0, 16.0)</td>
</tr>
<tr>
<td><strong>BMI (kg/m2)</strong></td>
<td></td>
<td></td>
<td>21.8(19.1, 26.1)</td>
<td>21.9(19.5, 26.0)</td>
</tr>
<tr>
<td><strong>Creatinine (mg/dL)</strong></td>
<td></td>
<td></td>
<td>168(114, 230)</td>
<td>139(83.0, 204)</td>
</tr>
<tr>
<td><strong>Cotinine (ng/ml)</strong></td>
<td></td>
<td></td>
<td>0.12(0.04, 0.79)</td>
<td>0.09(0.04, 0.64)</td>
</tr>
<tr>
<td><strong>T4 (ug/dL)</strong></td>
<td></td>
<td></td>
<td>1.49(0.99, 1.99)</td>
<td>1.49(0.99, 1.99)</td>
</tr>
<tr>
<td><strong>TSH (mIU/L)</strong></td>
<td></td>
<td></td>
<td>2.99(1.41, 5.61)</td>
<td>2.47(1.14, 5.37)</td>
</tr>
<tr>
<td><strong>Race:</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mexican American</td>
<td></td>
<td></td>
<td>185(8.3)</td>
<td>196(9.2)</td>
</tr>
<tr>
<td>Non-Hispanic White</td>
<td></td>
<td></td>
<td>131(5.9)</td>
<td>145(6.6)</td>
</tr>
<tr>
<td>Non-Hispanic Black</td>
<td></td>
<td></td>
<td>151(8.2)</td>
<td>164(9.1)</td>
</tr>
<tr>
<td>Other Race</td>
<td></td>
<td></td>
<td>39(8.0)</td>
<td>45(8.9)</td>
</tr>
<tr>
<td><strong>Menopause:</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td></td>
<td></td>
<td>NA</td>
<td>547(24)</td>
</tr>
<tr>
<td>Yes</td>
<td></td>
<td></td>
<td>NA</td>
<td>18(7.9)</td>
</tr>
<tr>
<td><strong>Alcohol Use:</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No alcohol</td>
<td></td>
<td></td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Less than a year</td>
<td></td>
<td></td>
<td>64(15)</td>
<td>105(24)</td>
</tr>
<tr>
<td>More than a year</td>
<td></td>
<td></td>
<td>153(25)</td>
<td>166(29)</td>
</tr>
<tr>
<td><strong>Income:</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$0–19,999</td>
<td></td>
<td></td>
<td>310(6.3)</td>
<td>358(7.0)</td>
</tr>
<tr>
<td>$20,000+</td>
<td></td>
<td></td>
<td>138(8.3)</td>
<td>143(8.7)</td>
</tr>
</tbody>
</table>
NA=Not Applicable; BMI=Body Mass Index; TCPY=3,5,6-trichloro-2-pyridinol

*a Frequency calculated by age*gender and represents percentage for male population in comparison to female population by age

*b Geomean and geometric standard deviation provided
### Table 2

Percent change\(^a\) in serum T4 (ug/dL) and TSH (mIU/L) levels associated with an IQR increase in urinary TCPY (ug/L) concentration

<table>
<thead>
<tr>
<th>Age group</th>
<th>12 – &lt;18 years</th>
<th>18 – 40 years</th>
<th>40 – 60 years</th>
<th>&gt;60 years</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Males</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>T4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Crude</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>2.00 (−0.58, 4.58)</td>
<td>2.83 (0.17, 5.49)</td>
<td>−0.51 (−3.40, 2.39)</td>
<td>−1.80 (−6.04, 2.43)</td>
</tr>
<tr>
<td></td>
<td>Adjusted(^b)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>3.85 (0.75, 6.95)</td>
<td>3.54 (0.13, 6.96)</td>
<td>−1.29 (−5.0, 2.42)</td>
<td>−1.62 (−7.77, 4.53)</td>
</tr>
<tr>
<td></td>
<td>TSH</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Crude</td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>0.38 (−7.36, 8.78)</td>
<td>−4.34 (−12.5, 4.57)</td>
<td>6.81 (−2.89, 17.5)</td>
<td>0.75 (−13.4, 17.3)</td>
</tr>
<tr>
<td></td>
<td>Adjusted(^b)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>4.73 (−3.91, 14.1)</td>
<td>−6.45 (−17.0, 5.48)</td>
<td>9.03 (−3.51, 23.2)</td>
<td>−14.5 (−27.6, −1.11)</td>
</tr>
<tr>
<td><strong>Females</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>T4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Crude</td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>−0.63 (−3.20, 1.94)</td>
<td>0.87 (−2.34, 4.09)</td>
<td>−1.66 (−5.74, 2.42)</td>
<td>3.57 (−1.26, 8.41)</td>
</tr>
<tr>
<td></td>
<td>Adjusted(^b)</td>
<td></td>
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<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>−0.07 (−3.26, 3.11)</td>
<td>−1.65 (−5.69, 2.39)</td>
<td>−2.44 (−7.09, 2.21)</td>
<td>−2.70 (−8.64, 3.25)</td>
</tr>
<tr>
<td></td>
<td>TSH</td>
<td></td>
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<tr>
<td></td>
<td>Crude</td>
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<td></td>
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<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>2.09 (−5.71, 10.4)</td>
<td>2.94 (−5.63, 12.3)</td>
<td>0.11 (−9.46, 10.7)</td>
<td>0.93 (−2.19, 22.9)</td>
</tr>
<tr>
<td></td>
<td>Adjusted(^b)</td>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
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<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>3.41 (−6.12, 13.9)</td>
<td>6.09 (−4.79, 18.4)</td>
<td>6.17 (−4.79, 18.4)</td>
<td>21.5 (3.37, 42.8)</td>
</tr>
<tr>
<td></td>
<td><strong>Weighted</strong></td>
<td></td>
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<tr>
<td></td>
<td>T4</td>
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<tr>
<td></td>
<td>Crude</td>
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</tr>
<tr>
<td></td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
<td>β (95% CI)</td>
</tr>
<tr>
<td></td>
<td>0.10 (−4.15, 4.35)</td>
<td>−1.15 (−5.39, 4.20)</td>
<td>−2.38 (−6.99, 3.69)</td>
<td>−1.65 (−6.99, 3.69)</td>
</tr>
<tr>
<td>Age group</td>
<td>12 – &lt;18 years</td>
<td>18 –40 years</td>
<td>40 – 60 years</td>
<td>&gt;60 years</td>
</tr>
<tr>
<td>-----------------</td>
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</tr>
<tr>
<td><strong>TSH</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td><strong>Crude</strong></td>
<td>9.19(−2.86, 22.7)</td>
<td>3.18(−5.94, 13.2)</td>
<td>5.90(−4.01, 16.8)</td>
<td>10.7(−2.96, 26.3)</td>
</tr>
<tr>
<td><strong>Adjusted</strong></td>
<td>7.30(−6.65, 23.3)</td>
<td>3.94(−8.06, 17.5)</td>
<td>10.9(−2.27, 25.8)</td>
<td>10.3(−3.57, 26.1)</td>
</tr>
</tbody>
</table>

*a* For T4, percent change relative to population median level.

*b* Adjusted for urinary creatinine, serum cotinine, BMI, age, race, income

*c* $p < 0.05